

Bioremediation 4.0

The Power of Procaryotic Bacteria Quorum Sensing and Signaling (QSS) & Organic Contaminant Destruction

TerraStryke Learning Program 23

Bacteria – History



Antoni van Leeuwenhoek 1632-1723



Founding Father of Microbiology



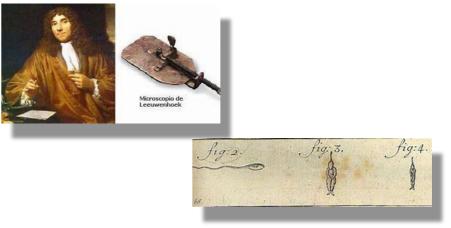
Identified planktonic bacteria - called them 'Animalcules' -



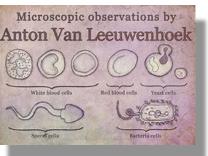
What he observed were protozoa, or 'little animals'.



Also observed, after time, test tube solution gelled.









Bacteria – History

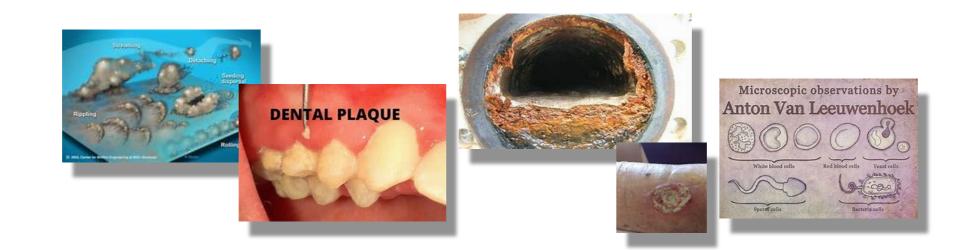


Late 1600's Leeuwenhoek noted biofilm bound bacteria.

Conveniently harvested from the plaque on his teeth.



Weren't aware of the complexity and prevalence of biofilm until the 1970s.





The Power of the Unicellular

Historically believed



Solitary

Capable of little

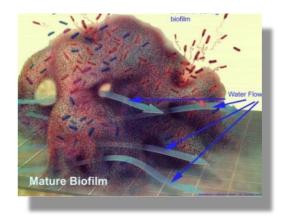
Swimmers key



We now have a completely different perspective



- < 1% of bacteria exist in planktonic form
- >99% of microbial populations live in biofilm
 - Communicate ('talk'), share information, and recruit
 - Determine what benefits the population
 - Abandon individual roles for specific roles
 - Establish structures like multi-cellular organism



What is a Biostimulation Strategy

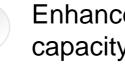
At TerraStryke, we wholeheartedly believe that the TREATMENT ZONE

needs to be viewed as an ecosystem that, WHEN CONTAMINATED, **IS UNDER DURESS**

and can not support healthy microbes or QSS.



#bioremediation4point0



Enhance geochemistry and growth capacity of treatment zone



Support indigenous populations



Restores nutritive capacity of ecosystem



Maximizes microbial information sharing

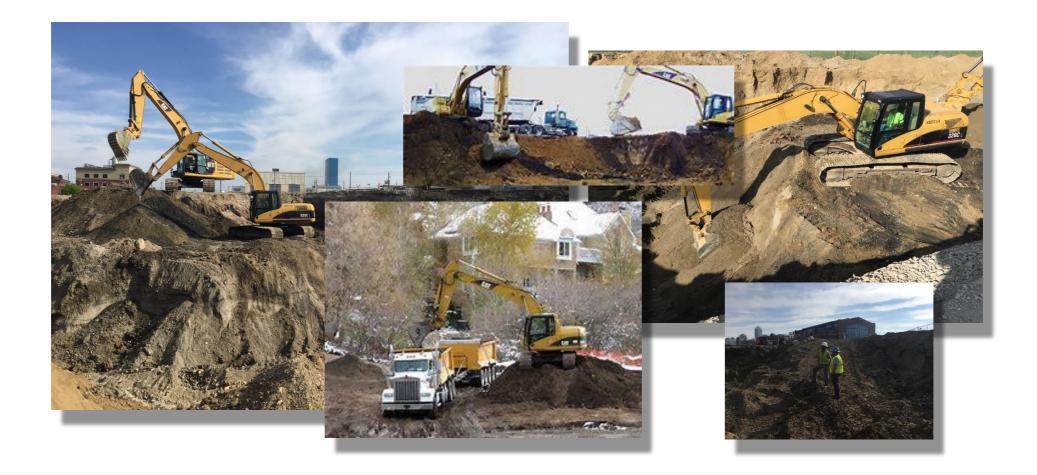


Establishes greater bulk fraction of contaminant degraders

O LIFE. TERRASTRYKE.COM GING THE GROUND

Biostimulation

What is not Biostimulation





Biostimulation

What is Biostimulation





Biostimulation

At TerraStryke, we wholeheartedly believe that the TREATMENT ZONE

needs to be viewed as an ecosystem that, WHEN CONTAMINATED, IS UNDER DURESS

and can not support healthy microbes or QSS.



#bioremediation4point0



Enhance nutritive capacity of treatment zone.



Support indigenous populations



Restores nutritive capacity of ecosystem.



Allows microbes to collectively establish biofilms.



Superior levels of sustainability, contaminant destruction with less impacts at lower costs.



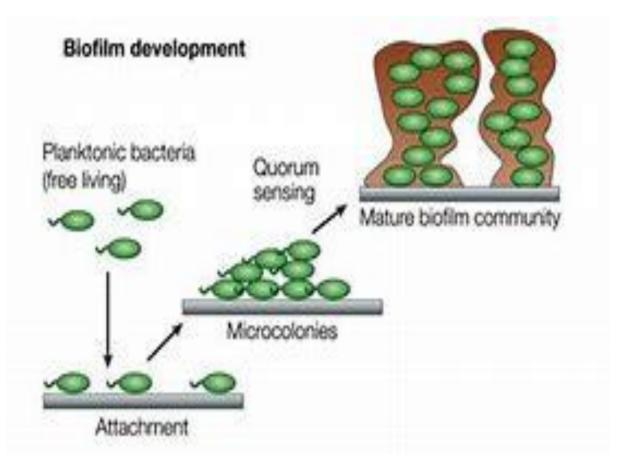
Most prevalent form of biology.



Biofilm

Biofilm development





Long-Chained Hydrocarbon Solution

TPHenhanced™ DESTROYS PHCs the way Mother Nature intended FOR LESS THAN YOUR DAILY YOUR DAILY CUP OF COFFEE ASK US HOW!

BUY NOW

TPHenhanced[™]

Passive-Aggressive Residual Contaminant Mass Destruction

Enhances respiration of heterotrophic bacteria under anaerobic conditions.

Organic strategy to contaminant destruction.

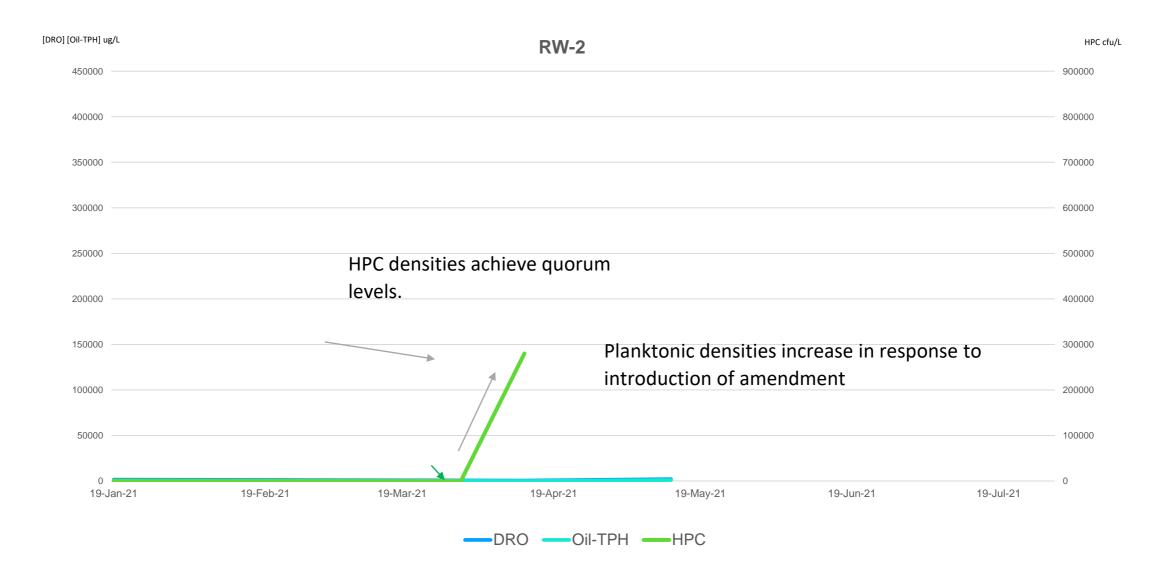
Proprietary macro-micro nutrient formulation package with nitrates to support ecosystem and respiration.

APPLICATIONS: Petroleum Hydrocarbons(PHCs) Polychlorinated Aromatics (PAHs) Naphthalene, MtBE, Creosoles. PCBs

Biostimulation supported PHC degradation in biofilm.



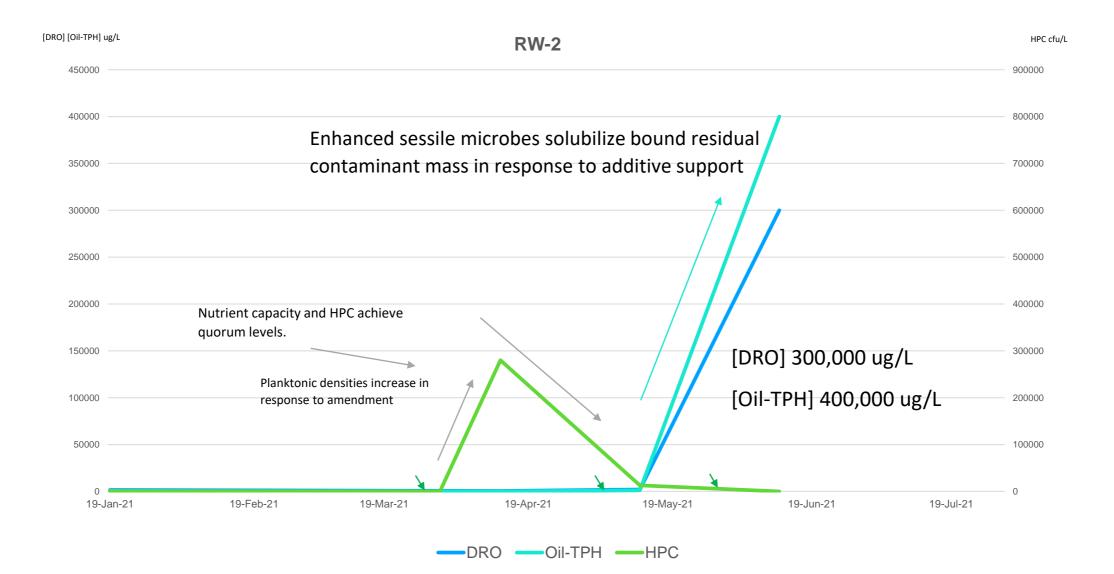
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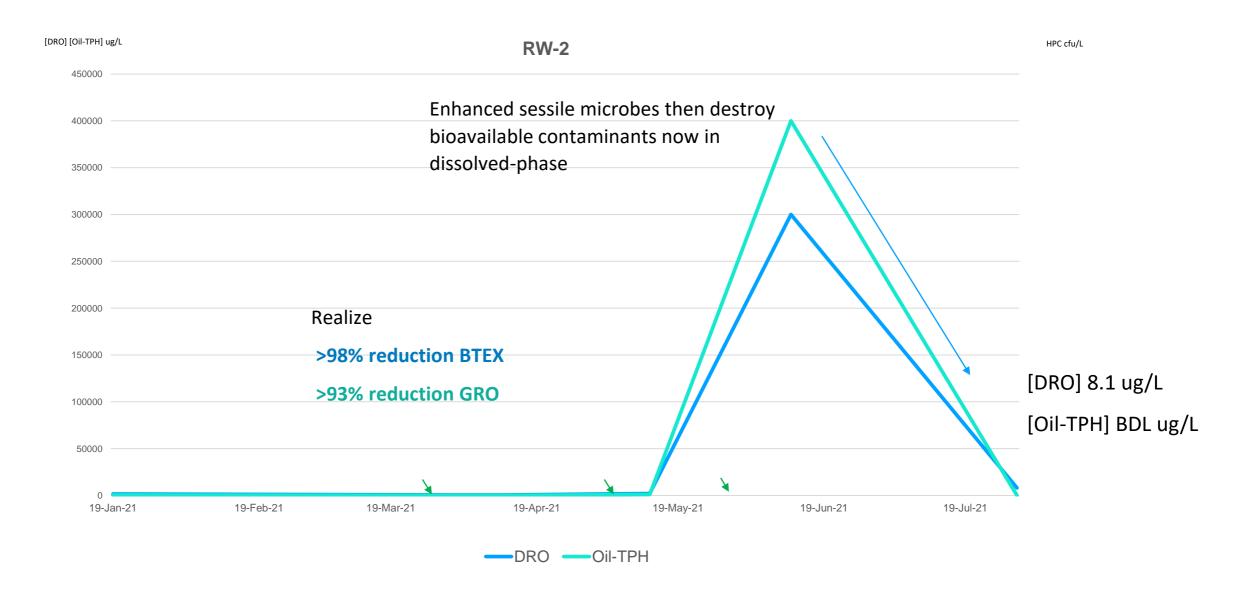
Biostimulation supported PHC degradation in biofilm.

[DRO] [Oil-TPH] ug/L		RW-2			HPC cfu,
450000					900000
400000					800000
350000			Microbes collect	tively respond wi	700000 ith
300000				nge from plankto	
250000	Nutriant conscituted UDC achieve sucre		HPC densities BI	DL	500000
200000	Nutrient capacity and HPC achieve quoru levels.	1111			400000
150000					300000
100000	Planktonic densities increase in response to amendment				200000
50000					100000
0 19-Jan-21	19-Feb-21 19-Mar-21	19-Apr-21	19-May-21	19-Jun-21	0 19-Jul-21
	-	DRO Oil-TPH	-HPC		

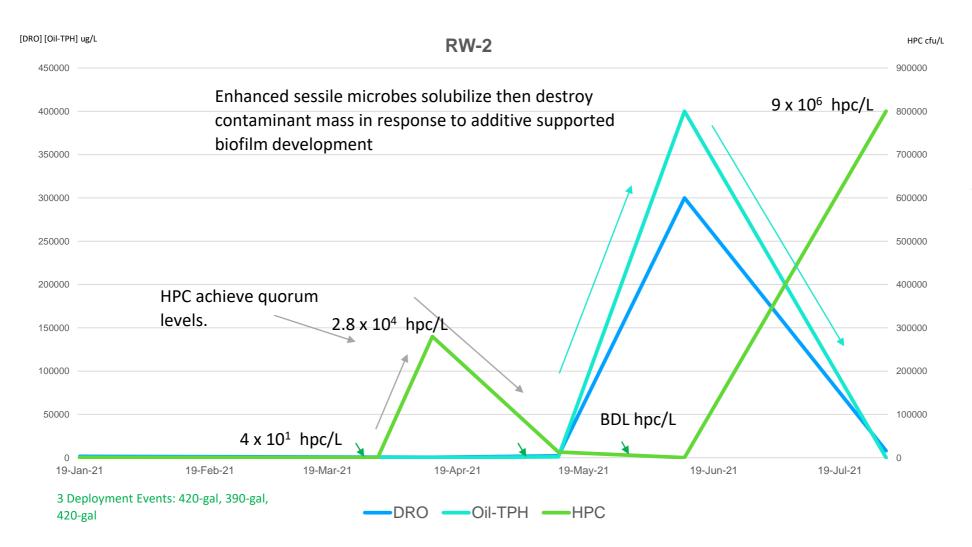
Biostimulation supported PHC degradation in biofilm.



Biostimulation supported PHC degradation in biofilm.



Biostimulation supported PHC degradation in biofilm.



Planktonic densities 1 month later increased to over 9,000,000 cf/L

Planktonic densities return only after >97%/>99.9% reductions from peak bioavailablility

Conclusions

TerraStryke biostimulation additives support the subsurface ecosystem and indigenous microbes to expedite:



LNAPL/DNAPL solubilization.



- Dissolved-phase contaminant utilization/destruction.
- The use of organic contaminants as electron donors/acceptors.



- Sustainable, climate neutral
- Sequester greenhouse gasses and eliminate aboveground support equipment



Simply by letting Nature have it.

WORKING TOGETHER, WE SUCCEED

Did you know that prokaryotic bacteria under suitable anaerobic conditions CHANGE PHENOTYPICALLY, COMMUNICATE/SIGNAL, BUILD, SHARE, AND WORK COLLECTIVELY?



#bioremediation4point0

Evaluation Amendments

ERDenhancedTM

Supports reducing conditions for decades after single injection program

APPLICATIONS: Dry cleaner, manufacturing, tool-dye

ERDenhanced[™]

SUSTAINABLE

cVOC remediation with complete destruction, without rebound,

- with NO multiple deployments
- with NO secondary contaminants
- with NO adverse affects

TERRA STRYKE

BUY NOW

Burlington, Ontario Site Former Dry Cleaner



Former Dry Cleaner

- [PCE] in saturated soil/groundwater
- Residual source mass in saturated soils



Site Conditions

- Highly weathered Shale with Silty-Sand
- Silt Generally moist
- 1-25ft bgs elevated PID readings

Property Value

• 2011 Appraised Value \$680,000









Burlington, Ontario Site Former Dry Cleaner



Excavation – Source Removal

- Excavated 250c.y. contaminated soils
- Infiltration gallery installed w/in footprint
- Clear stone, 6-inch slotted PVC, 6-9ft bgs



Additive Deployment

- Gravity fed 9% additive slurry
- 1,056 lbs to 1,100 gallons chase water March and again June 2014





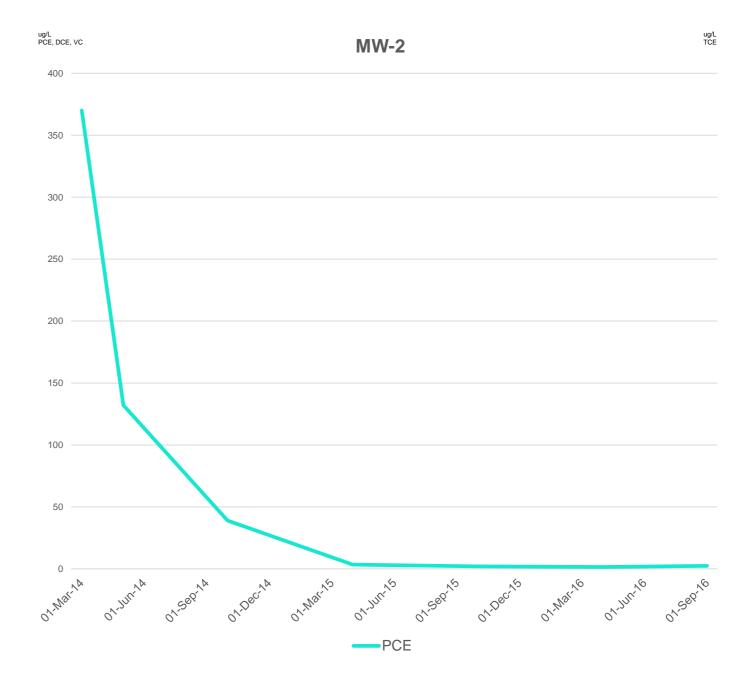


Burlington, Ontario Site Former Dry Cleaner

Results T=2 Years

MW-2 50-60ft downgradient

• 99.4% reduction [PCE]



Burlington, Ontario Site Former Dry Cleaner

Results T=2 Years

MW-2 50ft downgradient

- 99.4% reduction [PCE]
- After initial 32.1% increase
- 99.9% reduction [TCE] from peak.

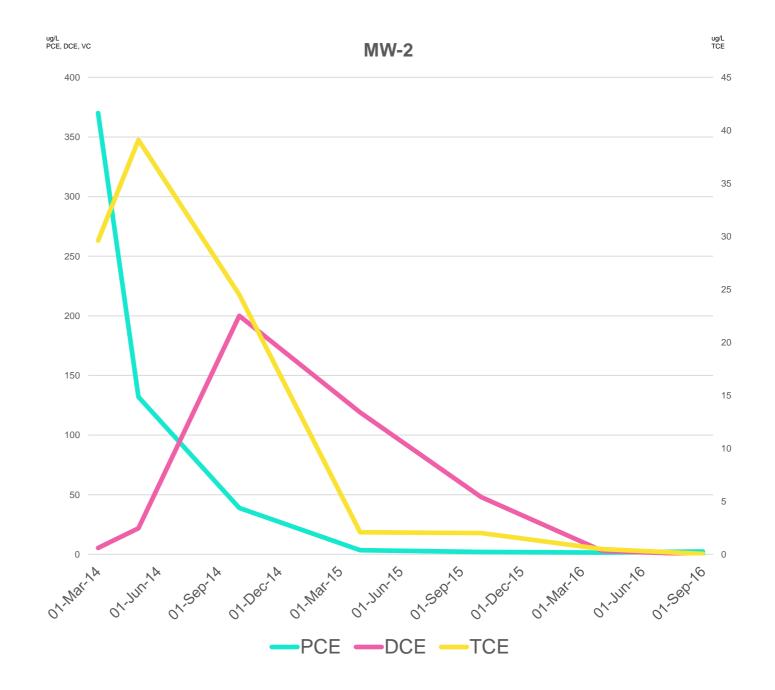


Burlington, Ontario Site Former Dry Cleaner

Results T=2 Years

MW-2 50ft downgradient

- 99.4% reduction [PCE]
- 99.9% reduction [TCE]
- After 3,600% increase
- >99.99% reduction [cis-DCE] from peak

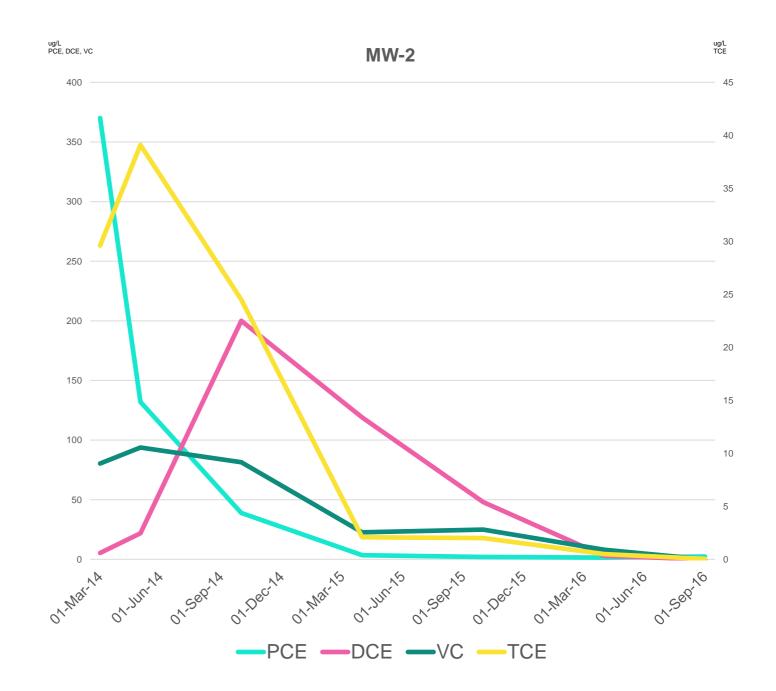


Burlington, Ontario Site Former Dry Cleaner

Results T=2 Years

MW-2 50ft downgradient

- 99.4% reduction [PCE]
- 99.9% reduction [TCE]
- ≈100% reduction [cis-DCE]
- 99.9% reduction [VC] after 16.8%↑
- 99.5% reduction in [cVOCtotal]
- [Ethene] generated throughout program = complete biotransformation



Burlington, Ontario Site Former Dry Cleaner



Pump & Treat? Property value \$680,000 P&T ≈ \$750,000-\$1M over 12-15 years Effective Property Value for 15-years \$0.00



Biostimulation Strategy

I	otal	project Costs	

Soil removal/gallery install	\$38,000
Pilot and Full-Scale Additive	\$35,000
Consulting and Analytical	\$150,000
	\$223,000



During 4th year of remediation Site redeveloped



Property Manager attributes \$1 million of property value increase to remediation strategy

2018 Property Value Assessed at MORE THAN \$2.5 million



28-day microcosm study

ERDenhanced alone

Biology outperformed

Baseline [PCE]

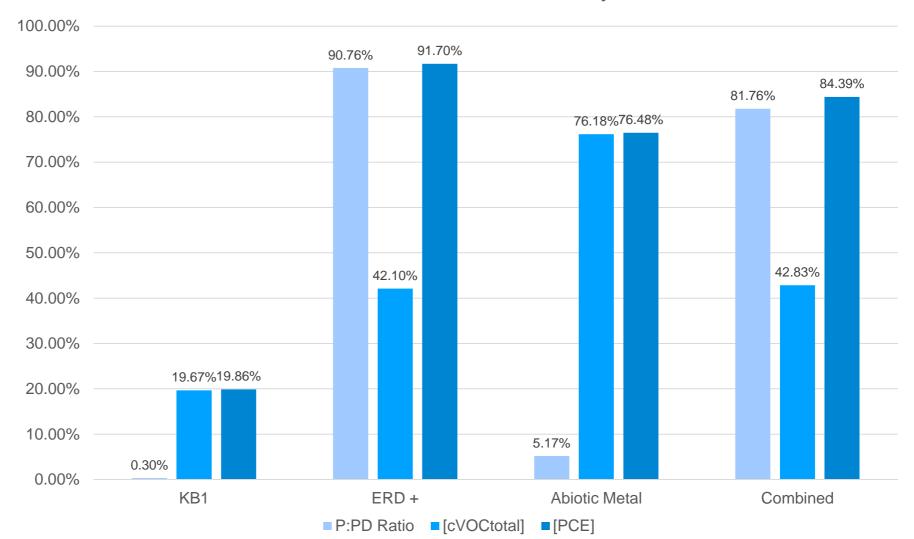
50,000 ug/L

best

abiotic

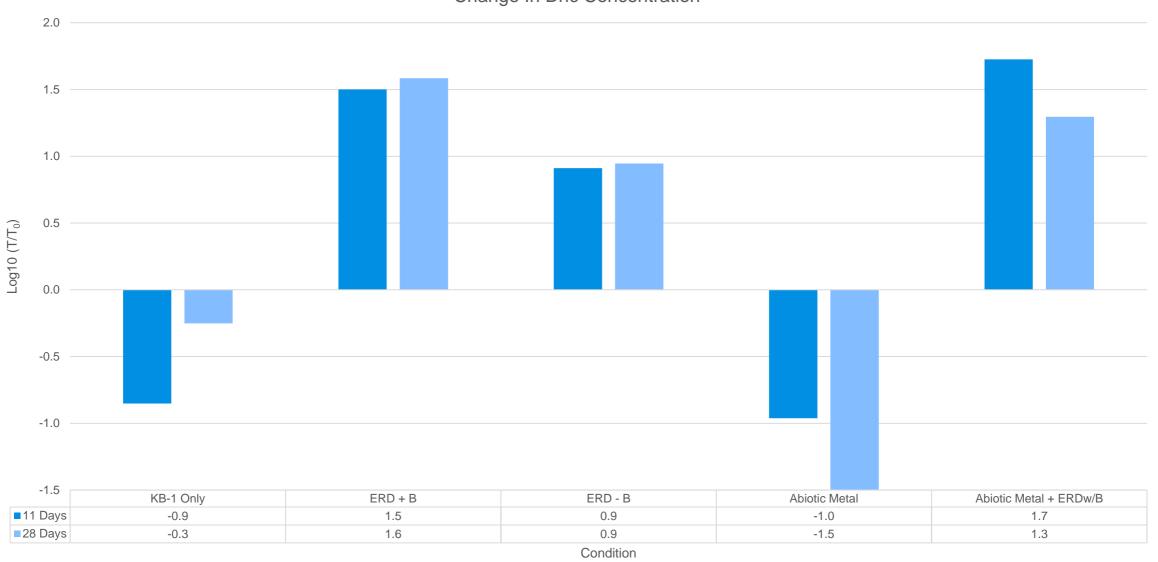
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Independent Comparative Evaluation



Percent Decreases cVOCs Day 28

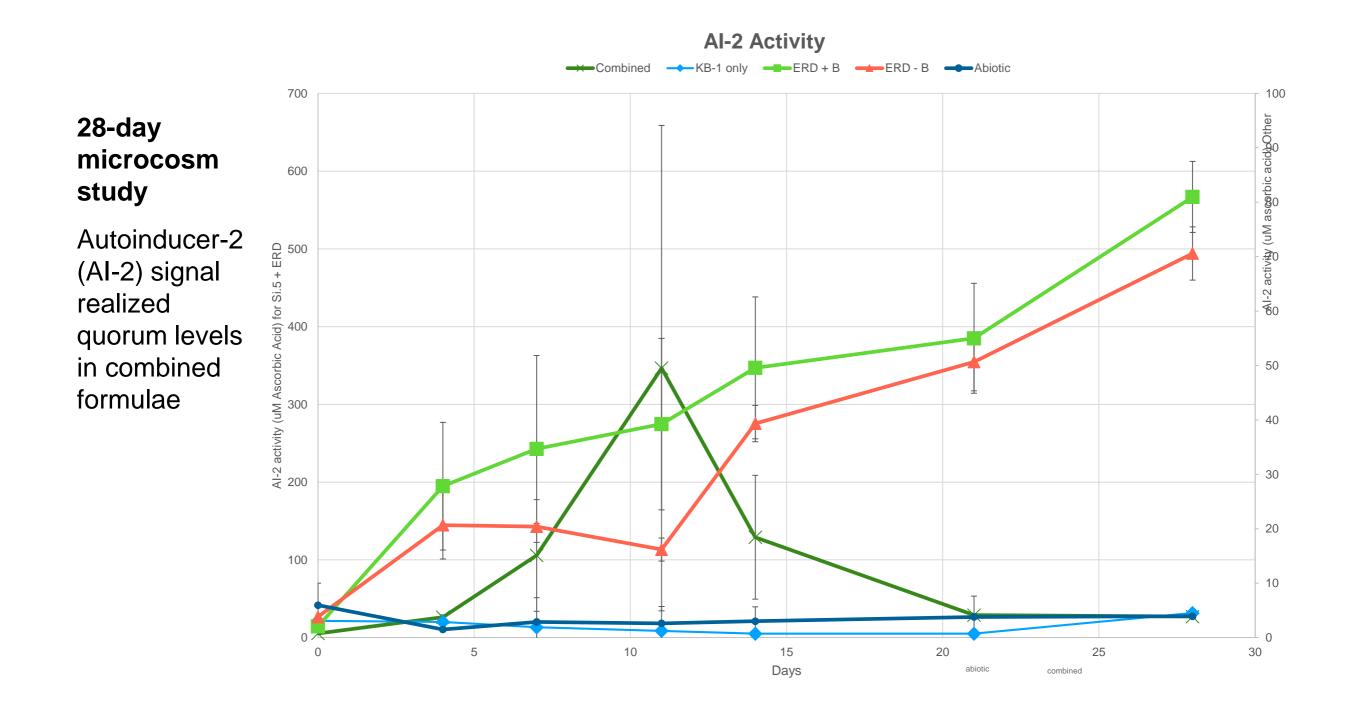
Independent Comparative Evaluation



Change In Dhc Concentration

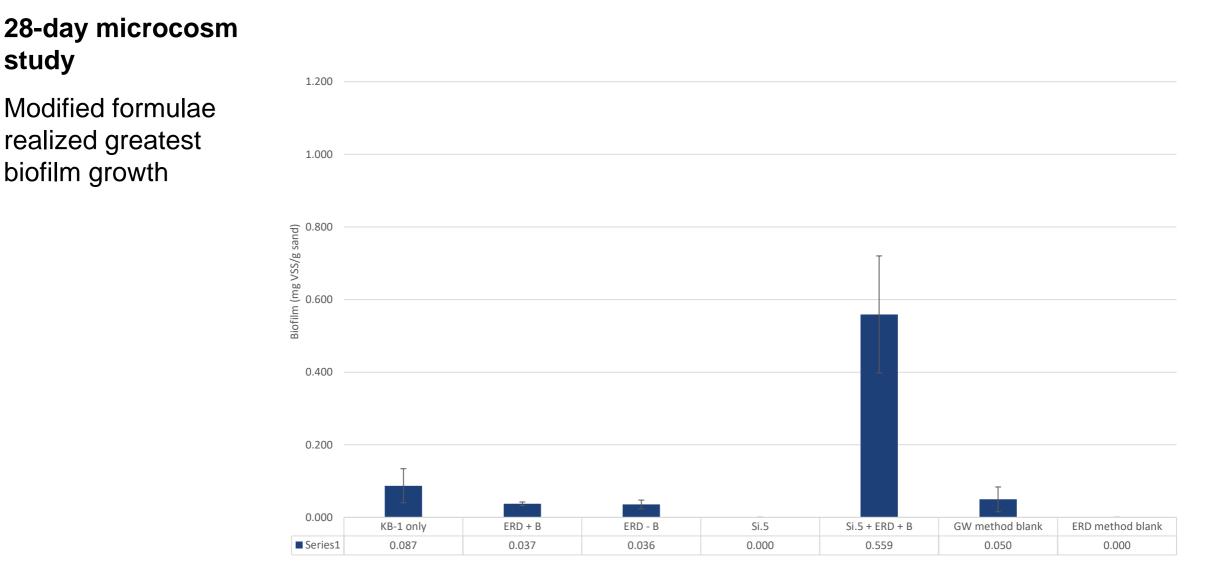
■11 Days ■28 Days

Independent Comparative Evaluation



Independent Comparative Evaluation

Biofilm Quantity



abiotic combined

Field Evaluation Study

Bridgeport Ohio

Former Electronics Manufacturing Facility

- Site[TCE] in saturated shallow bedrock
- 15-20ft alluvial silty clay/gravelly sand atop bedrock
- Bedrock highly fractured sandstone
- Residual DNAPL in 1%20 pore space of bedrock



Concern

- [TCE] 55-550 milligrams per Litre (mg/L)
- Minimal daughter product present
- Current P&T System manages plume migration



Goal

• Owner desires sustainable low-impact, low-cost strategy to target the destruction of dissolved phase and residual source mass contaminants.





Field Evaluation Study

Bridgeport Ohio

Former Electronics Manufacturing Facility



Strategy

- On-Site proof-of-concept evaluation
- Performed under actual biogeochemical conditions
- Compared ERDenhanced standard formulation
- to modified version containing a minimal % of electron generating metal



Process

- Amend monitoring/test wells using Passive Release Sock (PRS)
- One with original, one with modified ERDenhanced
- Monitor/sample test wells over 12-month evaluation

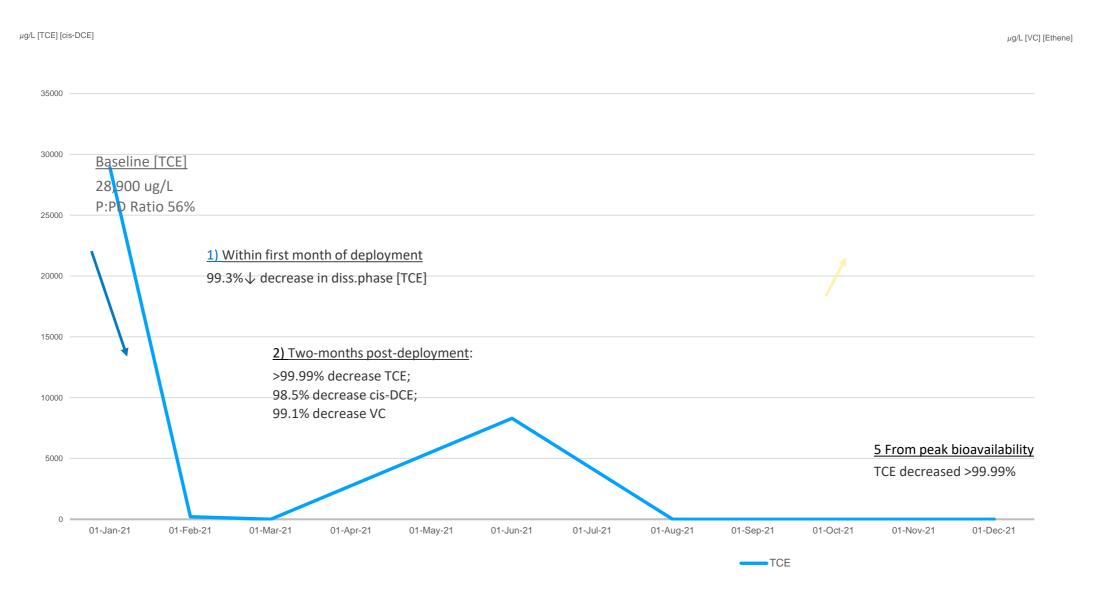


Goal

- Realize increased densities of indigenous microbials
- Expedited residual mass solubilization
- Enhanced and complete dissolved phase cVOC destruction

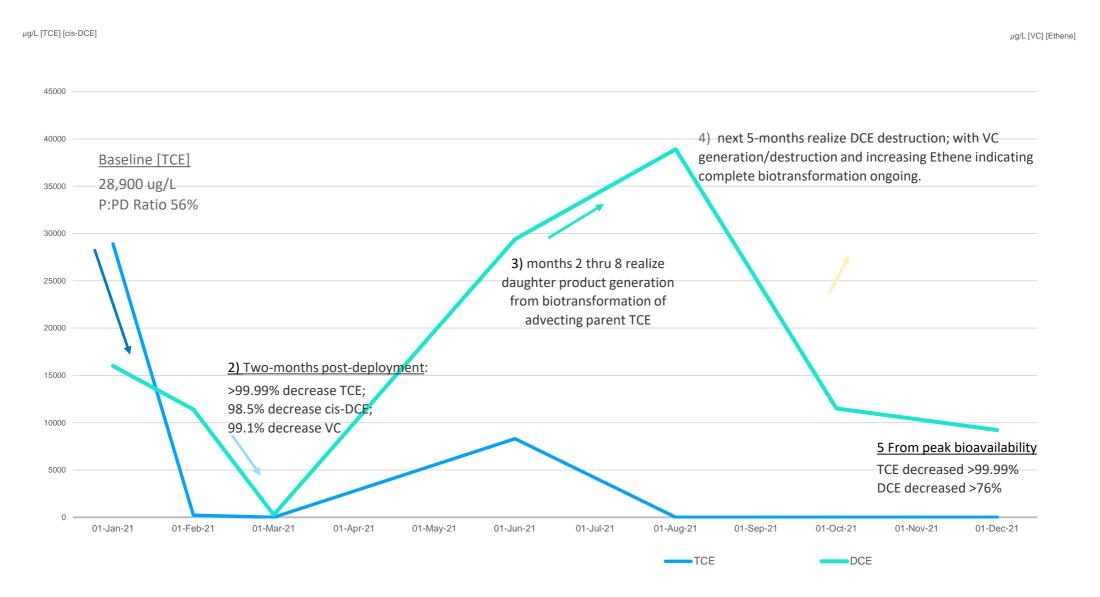
On-Site Proof-of-Concept

ERDENHANCED MW-23A 12-month evaluation



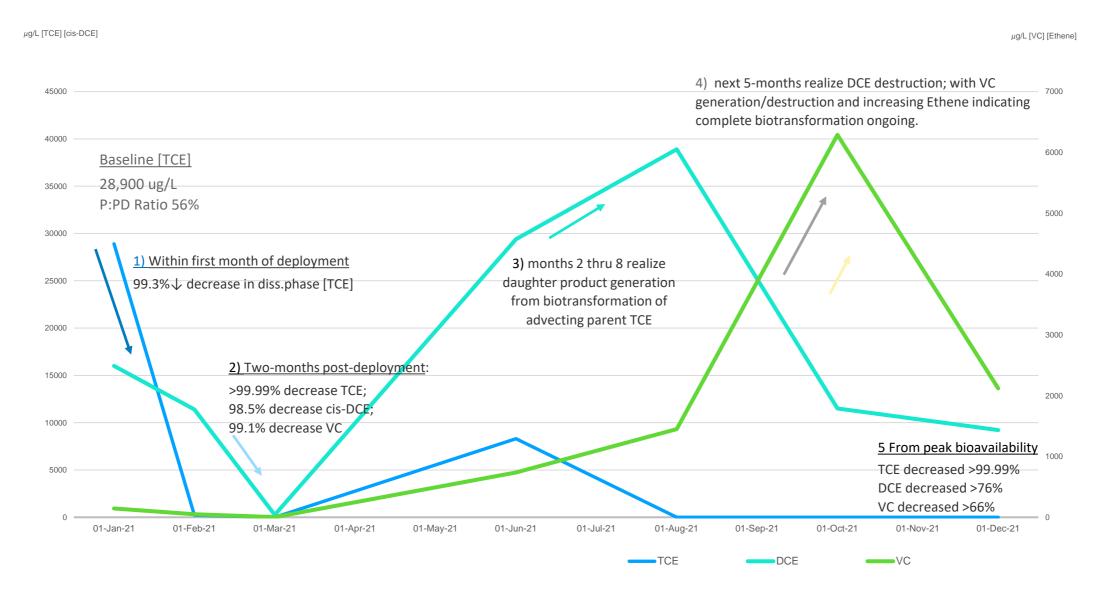
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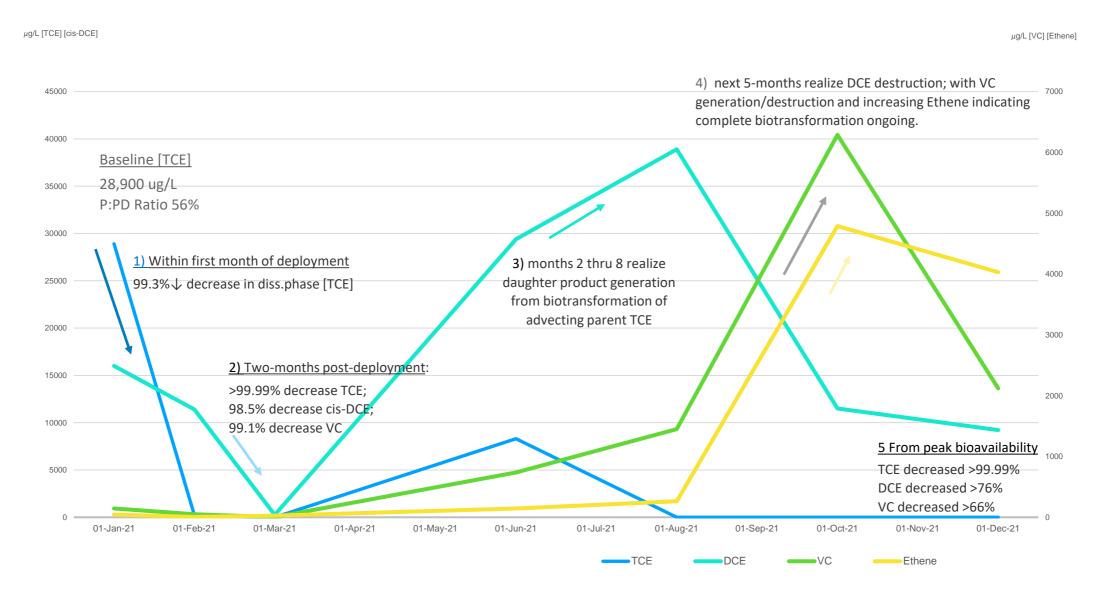
On-Site Proof-of-Concept

ERDENHANCED MW-23A 12-month evaluation



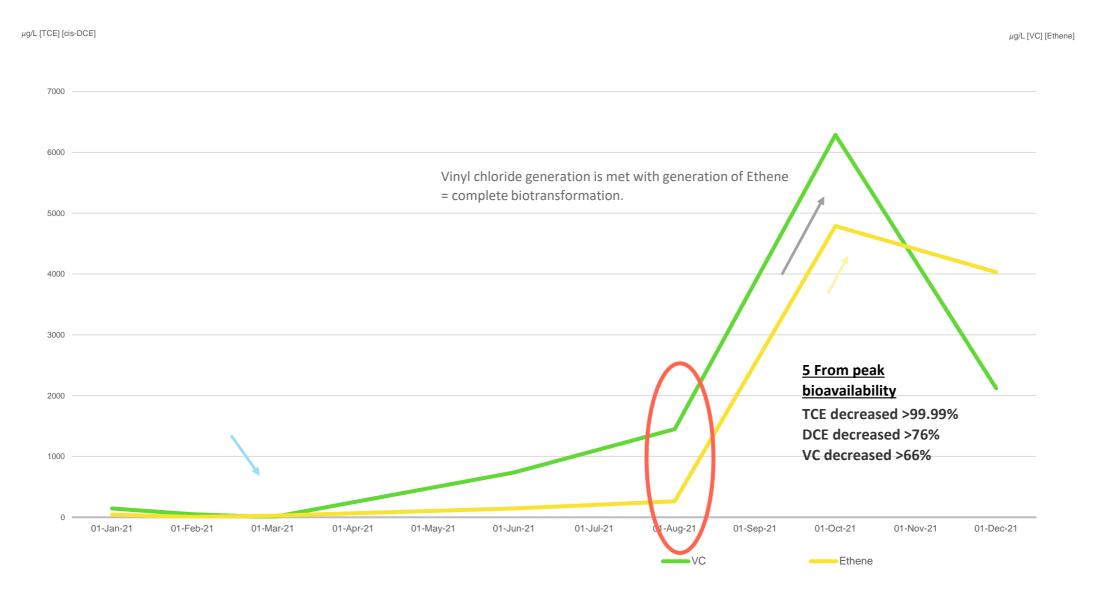
On-Site Proof-of-Concept

ERDENHANCED MW-23A 12-month evaluation



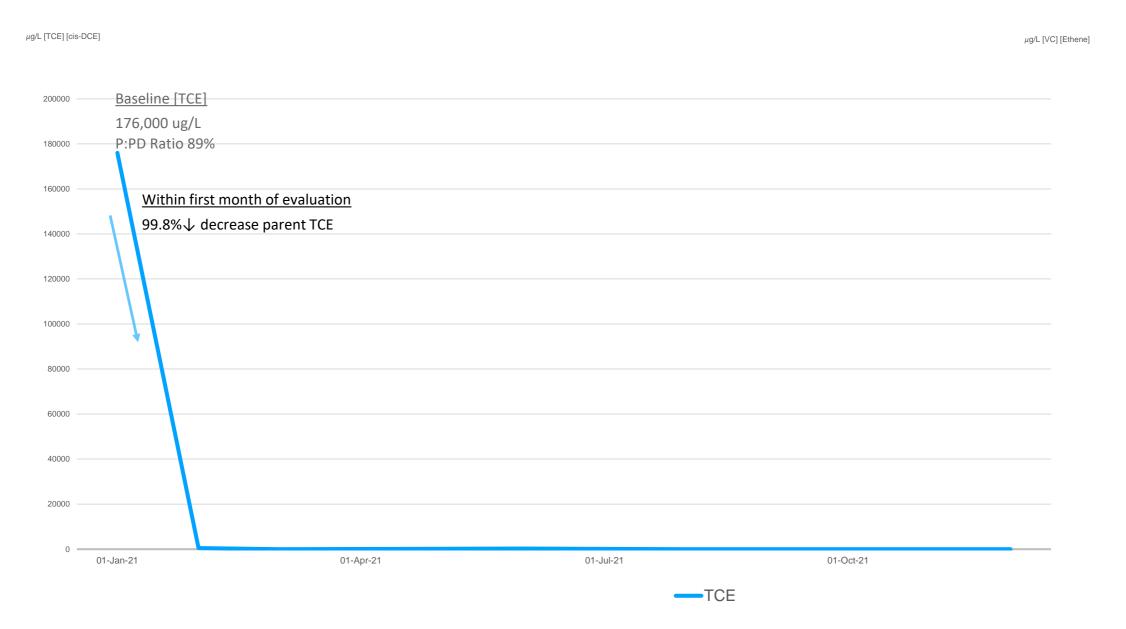
On-Site Proof-of-Concept

ERDENHANCED MW-23A 12-month evaluation



On-Site Proof-of-Concept

ERDENHANCED W/ZVI MW-24A



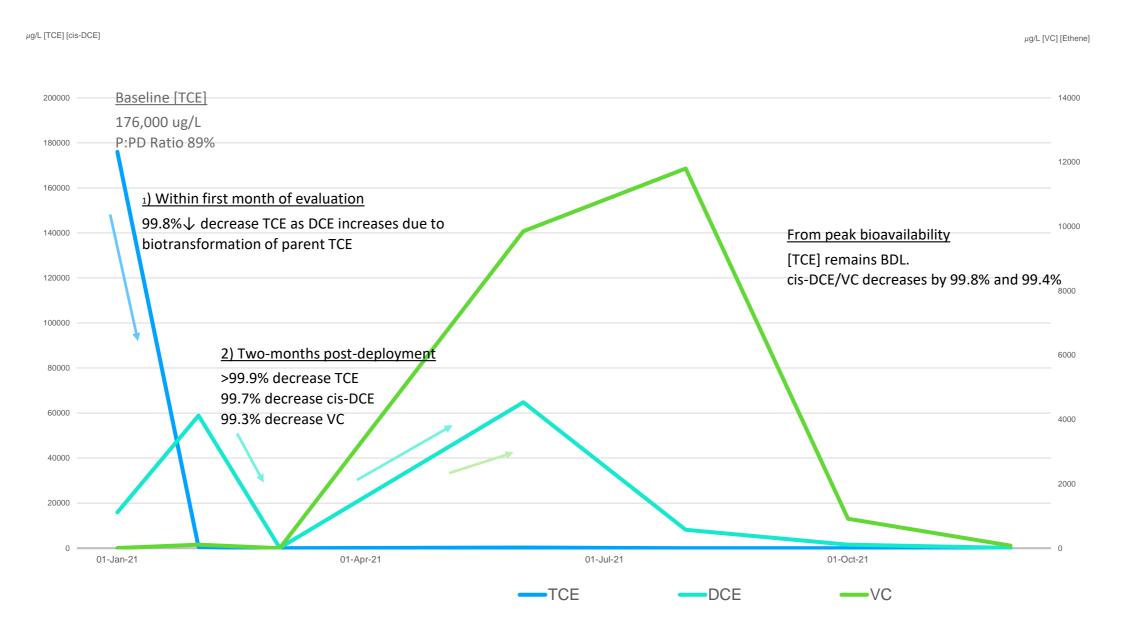
On-Site Proof-of-Concept

ERDENHANCED W/ ZVI MW-24A

µg/L [TCE] [cis-DCE] µg/L [VC] [Ethene] Baseline [TCE] 200000 176,000 ug/L P:PD Ratio 89% 180000 160000 1) Within first month of evaluation 99.8% \downarrow decrease TCE as DCE increases due to 140000 biotransformation of parent TCE 120000 100000 Two-months post-deployment 80000 >99.9% decrease TCE 99.7% decrease cis-DCE 60000 40000 20000 0 01-Jan-21 01-Apr-21 01-Jul-21 01-Oct-21 -TCE DCE

On-Site Proof-of-Concept

ERDENHANCED W/ ZVI MW-24A



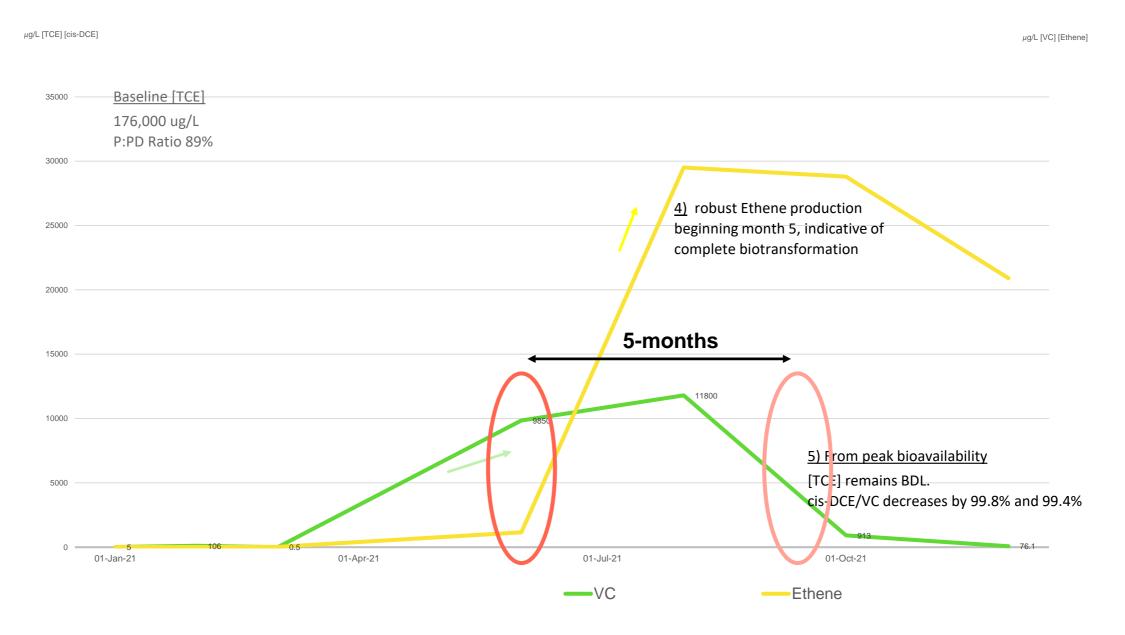
On-Site Proof-of-Concept

Modified ERDENHANCED MW-24A

µg/L [TCE] [cis-DCE] µg/L [VC] [Ethene] 200000 Baseline [TCE] 35000 176,000 ug/L P:PD Ratio 89% 180000 30000 160000 1) Within first month of evaluation 4) robust Ethene production 99.8% \downarrow decrease TCE as DCE increases due to beginning month 5, indicative of 25000 140000 biotransformation of parent TCE complete biotransformation 120000 20000 100000 3) DCE/VC daughter production less 2) Two-months post-deployment 15000 significant than MW-23A; faster 80000 >99.9% decrease TCE biotransformation? 99.7% decrease cis-DCE 60000 99.3% decrease VC 10000 From peak bioavailability 40000 [TCE] remains BDL. 5000 cis-DCE/VC decreases by 99.8% and 99.4% 20000 0 01-Jan-21 01-Apr-21 01-Jul-21 01-Oct-21 -TCE -DCE -VC Ethene

On-Site Proof-of-Concept

ERDENHANCED W/ZVI MW-24A



Field Evaluation Study

Summary Former Electronics Manufacturing Facility



ERDenhanced

- Achieves robust and complete DNAPL dehalorespiration
- Without 'cis-stall', sustainably for decades after single deployment



Modified Formulation

- Minimal additional of electron generating metal
- Catalyzes biology
- Expedites diss.phase contaminant reductions; while,
- Realizing complete and long-term dehalorespiration of DNAPL and diss.phase contaminants

Conclusions

TerraStryke biostimulation additives support the subsurface ecosystem and microbes to expedite:



LNAPL/DNAPL solubilization.

Dissolved-phase contaminant utilization/destruction.

The use of organic contaminants as electron donors/acceptors.



Achieve sustainable remediation without above ground equipment costs/permitting.

Sequester Greenhouse Gasses.



Realize Site Compliance with less impacts, less costs simply by letting Nature have it.

WORKING TOGETHER, WE SUCCEED

Did you know that prokaryotic bacteria under suitable anaerobic conditions CHANGE PHENOTYPICALLY, COMMUNICATE/SIGNAL, BUILD, SHARE, AND WORK COLLECTIVELY?



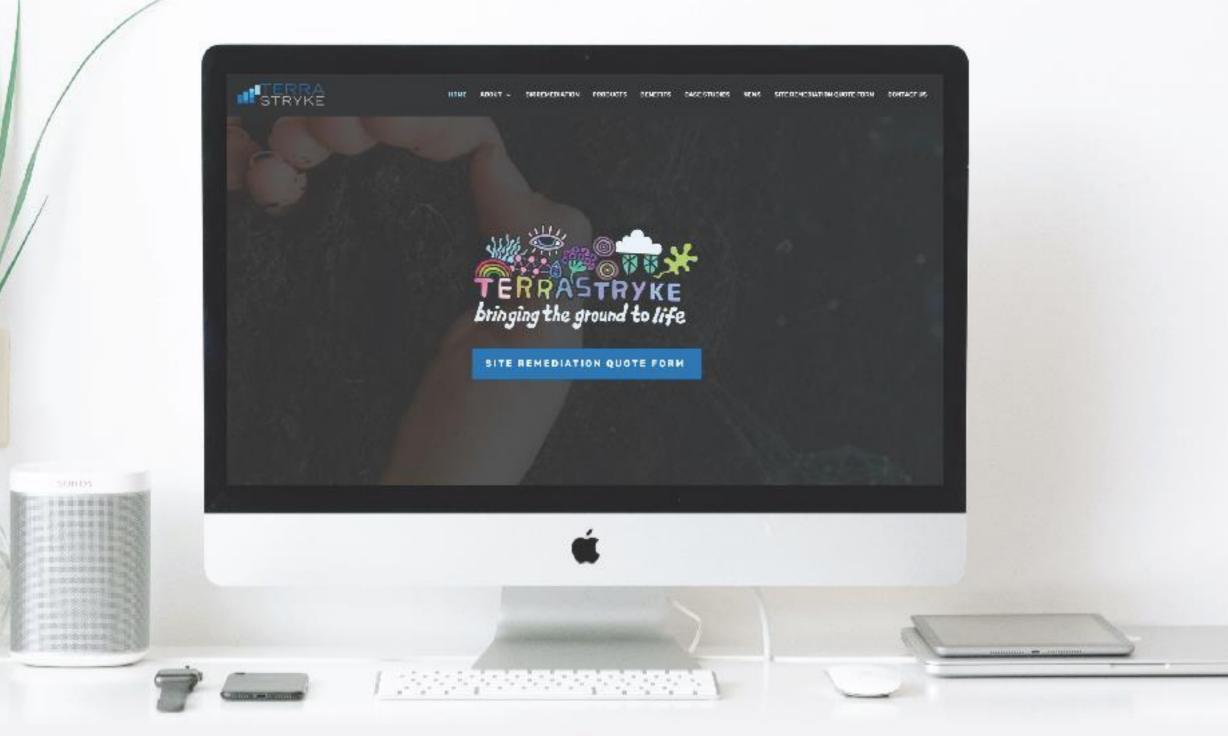
#bioremediation4point0

Conclusions

There are lots of options out there



Site Remediation Quote Form



Contact Information

IF YOU HAVE A CONTAMINATED SITE THAT NEEDS CLEANING UP, REACH OUT TO US!



Kent C. Armstrong President

(603) 977-0810 (office) (603) 731-3159 (cell) karmstrong@terrastryke.com

284 Depot Street / P.O. Box 254 Andover, New Hampshire USA

950 Fennell Avenue, Suite 105 Hamilton, Ontario CDN L8V 1X2 (905) 387-2255

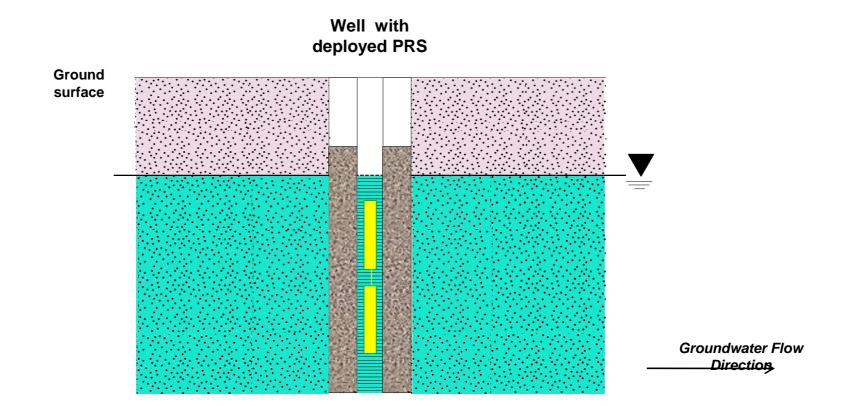


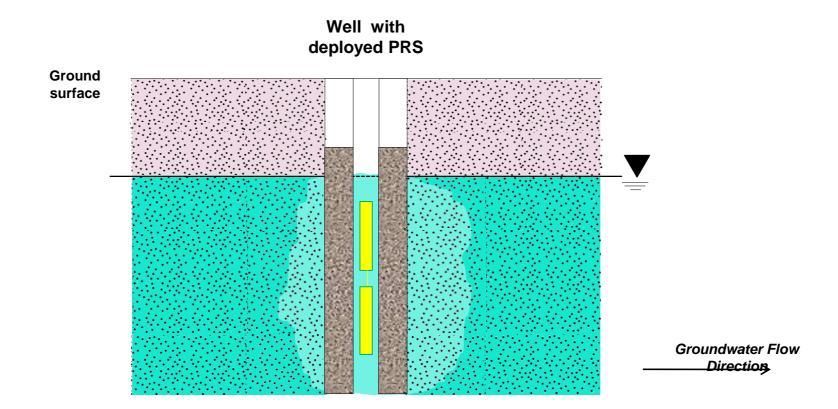
Passive Release Sock (PRS Deployment Unit)

- Low-Cost, Low-Risk On-site Evaluation process
- Fit directly into existing 2-inch GW monitoring well
- Provides Representative 'Go-no-Go' on-Site Evaluation
- Baseline & Performance Monitoring/Sampling
- Field Indicator Parameters Recorded Every Replacement Event
- ORP, DO, pH, Temp, Cond; NO₃, SO₄, dissolved Mn/Fe; Ethane, Methane, Ethene, and Contaminant of Concern
- Non-purge, low-flow sampling protocols

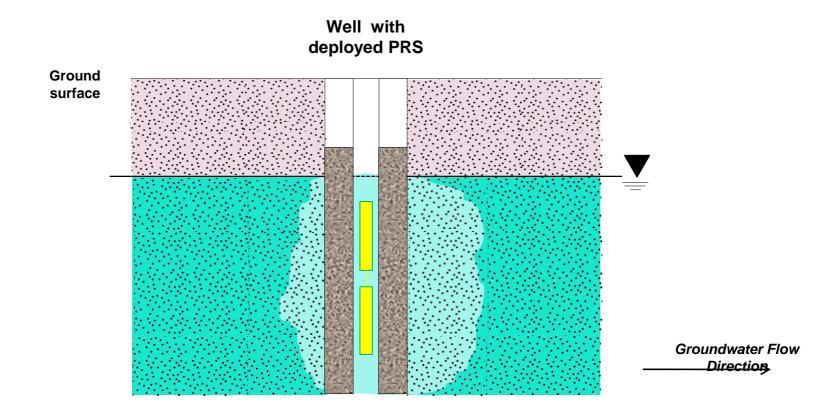




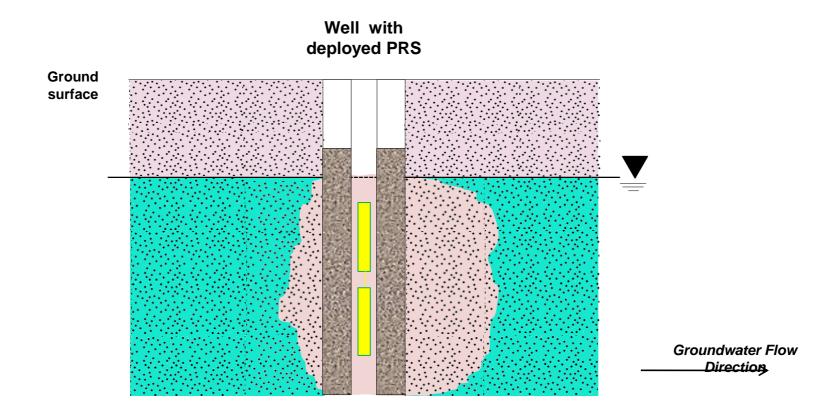


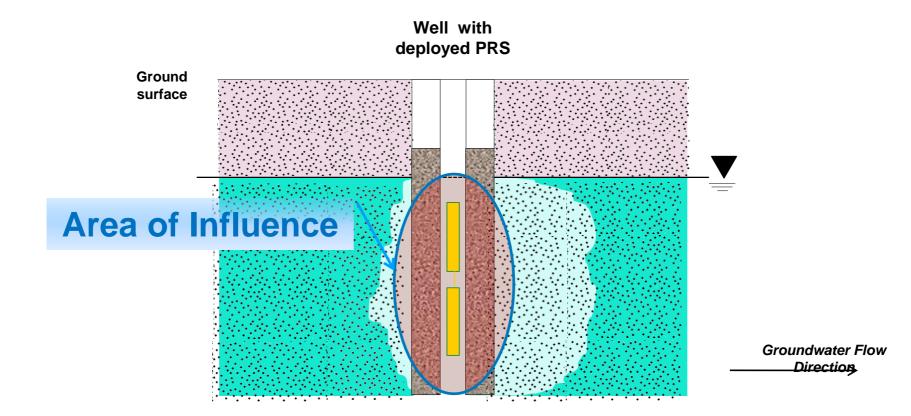






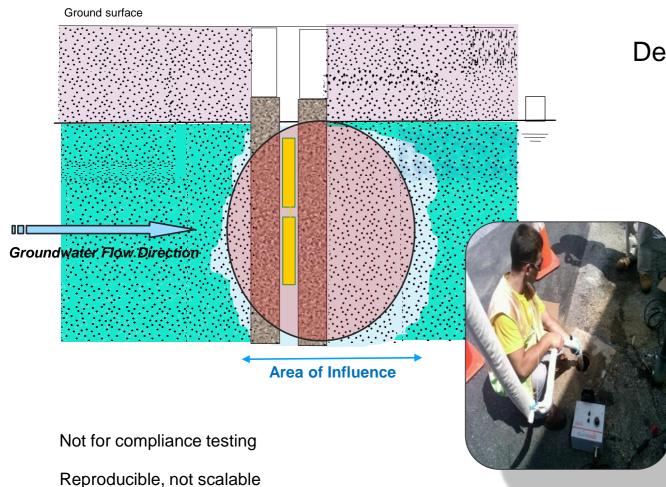








PRS Pilot Study Schematic



'Go-no-Go' evaluation Additive filled Passive Release Sock (PRS) Deployed into existing 2-inch gw monitoring well

> Passively amend saturated screened interval Create 1-2 meter area-of-influence Replace PRS units every 6-8 weeks

Monitoring Program

Baseline Each replacement event Non-purge Low-flow 6-8 replacement events typical